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SOME CONSIDERATIONS OF PROTOPLASM.

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One of the most fundamental problems in biological science is that which concerns protoplasm. Yet there is great diversity of opinion among biologists regarding what constitutes protoplasm and some doubt whether the term protoplasm is really worth retaining. In the present state of knowledge, protoplasm can not be defined in any terms of physical structure which will be accepted, without qualification, by a majority of botanists, and can only be defined somewhat more satisfactorily in terms of colloidal chemistry. Again, though chemical definition is somewhat more certain than physical, this alone is far from satisfactory to those who think of cell contents in terms of microscopic structure.

It is sometimes stated by certain biologists that protoplasm is essentially alike in all organism. This may be true in the rough if we define in purely chemical terms; or if we content ourselves with the statement that protoplasm is the living substance of the cell, knowing not how much of the cell is alive, and, therefore, protoplasm. Turning to those very lowly organized plants, the bacteria, most of us will agree that the whole cell content, inclusive or exclusive of the vacuoles, composes the protoplasm. For higher fungi and for animals the situation is about the same, except that definite nuclei here replace the nuclear granules commonly supposed to exist in bacteria. Turning attention to higher green plants, we find that the cells are much more complex with respect to visible contents. This is especially true of all the cells of lower algae and of those cells of higher bryophytes, pteridophytes, and spermatophytes in which photosynthesis is carried on.

Regarding these chlorophyllous, photosynthesizing cells of green plants, every possible position has been taken respecting the portion of visible cell contents which may constitute the protoplasm.

Knowing well that the problem was not a very fundamental one, the writer, like other teachers and investigators, has had to puzzle about what visible and therefore obstrusive portions of the protoplast or whole cell content should be regarded as protoplasm. Finally, in November, 1923 he addressed a questionnaire to about sixty teachers in colleges and universities, merely asking which of these elements each one would regard parts of the protoplasm. More than forty replies resulted from the letters; and the views expressed were readily arranged in four general catagories for presentation at the meeting of the Botanical Society of America at Cincinnati, in December, 1923. Following this, those who had not replied and a few others were circularized, asking which of the four catagories which resulted from the first letters each one would favor. The result of the two efforts was more than sixty replies. The circular letters stated that the writer would feel free to quote from the replies; and quotations from some of the most valuable discussions will form an important portion of this paper. The statistical results might have been more valuable if a larger number of botanists had been consulted. Yet the selection of persons whom to address was carefully made; and the writer believes that what follows will be found valuable, especially to those who have had little time for the formulation of a basis for thinking and teaching regarding protoplasm.

Seven of the botanists who replied adhered to one position which grew out of the questionnaire, namely that protoplasm consists of cytoplasm and nucleoplasm, the latter more commonly in the form of a definite nucleus. The adherents to this belief are H. C. Cowles, R. P. Hibbard, E. C. Jeffrey, O. E. Jennings, C. B. Lipman, G. B. Rigg, and A. R. Sweetser. After a general discussion, Hibbard says "In many cases I would agree with class two, but there are some masses of nucleated protoplasm that do not contain plastids. So to cover all cases I would take my stand with class one." In explanation it should be stated that class two as presented at Cincinnati is the next following. Jeffrey says in part "I suppose, strictly speaking, the term protoplasm should be limited to the nitro-

genous, undifferentiated contents of cells. If one goes beyond that, I do not see how it is possible to avoid everything, including the wall." The present writer began many years ago with the belief held by these seven men, but turned recently to the view that the whole cell content should be regarded as protoplasm. However, the wall is like certain cell inclusions with respect to origin, and he sees plainly the force of Jeffrey's logic. The following from Jennings is peculiar: "Possibly plastids have as their original basis certain differentiated portions of the protoplasm, just as I would consider the nucleus a highly differentiated portion of the cytoplasm." Of course the relation of plastids and nuclei to protoplasm and cytoplasm depends in part on definition of these last two components of protoplasts, and the writer is content for the present to quote Jennings without further comment.

After stating his view, Cowles says "I have talked with Chamberlain, who gives our Cytology courses, and he also treats the subject as I have." It appears, however, that Chamberlain believes quite differently. He says "For convenience of reference, I use the terms cytoplasm and nucleus; but I do not use the term protoplasm to cover the two taken together. Whenever we use the term protoplasm at all, we use it loosely as a synonym for cytoplasm." Shull's statement, quoted in full later in this paper, differs from both Cowles and Chamberlain. There are advantages in likeness of presentation within a department; but of course independence in thinking is preferable. Coulter referred the writer to Shull as spokesman for the department; but it is stimulating to find that the department cannot be spoken for by one man.

According to the best analysis that the writer can make, thirty-eight botanists addressed took essentially a second position which grew out of the questionnaire, namely that protoplasm consists of cytoplasm, nucleoplasm and plastids. These botanists are G. M. Armstrong, H. W. Barre, H. H. Bartlett, A. R. Bechtel, D. H. Campbell, A. H. Chivers, H. S. Conard, M. T. Cook, O. F. Curtis, B. M. Davis, A. W. Evans, J. H. Faull, M. C. Ferguson, W. F. Ganong, W. A. Gardner, F. O. Grover, J. W. Harshberger, D. S. Johnson, F. D. Kern, J. E. Kirkwood, B. E. Livingston, J. N. Martin, H. F. A. Meier, F. T. McFarland, A. Nelson, S. P. Nichols, L. H. Pammel, R. J. Pool, J. H. Schaffner, W. A. Setchell, L. W. Sharp, C. A.

Shull, A. G. Stokey, E. N. Transeau, W. Trelease, A. E. Waller, K. M. Wiegand, and R. B. Wylie. Davis qualified by calling attention to the fact that plastids are not present in parasitic and saprophytic plants. Faull stated that "pyrenoids are looked upon as specialized areas within the plastids and constitute a part of the plastid substance." This statement is related to the addition of pyrenoids by one or two other botanists. Johnson added that blepharoplasts are sometimes to be included. Martin would add pyrenoids and chondriosomes. Sharp referred me to his text on Cytology, and it would seem from statements, pages 133 to 135, that he belongs with this group. Wylie would add to cytoplasm, nucleoplasm, and plastids the plasma membrane. Bartlett wrote at considerable length, and want of space compels doing injustice to his valuable statement by passing it with record of the fact that he regards water and chlorophyll parts of protoplasm.

Campbell's statement is much shorter, and it represents the general belief of these 38 botanists fairly well. His statement runs thus: "In discussing protoplasm before my classes, I have considered the whole living contents of the cell, cytoplasm, nucleus, and plastids as constituting the protoplasm. I should not include vacuoles, starch grains, protein granules, or cell wall under this head; and I have tried to make clear to students that we can not look upon protoplasm as a definite chemical substance, but that it is a mixture of a very great number of complex substances, and is in a state of constant change, due to the activities within. Hence no chemical analysis can represent the true composition of living protoplasm as it must also include various inclusions of the protoplast which are not constituents of the living protoplasm."

Livingston's 800-word statement deserves quoting as a whole, but we can take space only for the following, which contains an interesting thrust and several very illuminating comparisons. He says in part "I don't care what protoplasm is so long as we know how it is built and how it operates. The problem is somewhat like asking for a definition of soil or air. If I take the water out of the soil or air, I still have soil or air. If I take the nitrogen out of air, it is still air, I suppose, but perhaps it isn't if I take the oxygen out. Take the dissolved salts out of soil, or take the dissolved gases out of soil, and it is still soil. But take the solid grains out, and we don't consider

it soil any more, even though the undissolved gases are present, as in sea foam. It seems foolish to try to define air, or soil, or protoplasm, or mince-meat, or plum-pudding, or chocolate fudge, or house-paint. When we attempt to define them we discover that there are many kinds of each, and that all of the constituents of any one mixture may be left out and replaced by different materials, without requiring the use of another general term. If a whole apple were found in the middle of the plum-pudding, we'd not call it a part of the pudding any more than if it were the baby's rubber doll that had been included by mistake. But if we took all of the dead grapes out of the plum-pudding, it wouldn't be a plum-pudding any more! Apparently the distinction is based largely on size. Small pieces of apple are part of mince-meat, but larger pieces (fix your own limits of size) would be considered foreign bodies surrounded by mince-meat." It is from other portions of his discussion that it was concluded that Livingston believes in a general way that protoplasm consists of cytoplasm, nucleoplasm, and plastids; and some violence has been done in taking the above quotation out of its setting because of its intrinsic value.

Shull has expressed himself as follows: "For some time I have used the criterion of actively living substance for living protoplasm; and for dead protoplasm, such material as had been once actively living. I have never considered the vacuoles, starch grains or protein granules, or oil droplets as real parts of the protoplasm. They are parts of the cell when one is classifying the structures found in the cell unit of structure; but I would consider the starch grains and other food inclusions as mere storage products, stored temporarily and conveniently in the region of living substance for more or less immediate use. But I do not see how we could consider them parts of the living substance until they had been assimilated and synthesized into metastable protoplasm molecules of some kind. On the other hand there is reason for considering chloroplasts and leucoplasts as protoplasmic bodies, likewise mitochondria. Certainly a part of their substance is actively living, whether the chlorophyll is or not. From this point of view the wall would hardly be called living matter. It is no more alive than the starch grain, and like the starch grain may be considered the condensed product of enzymic action on the soluble carbohy-

drates. It seems to me highly probable that the pyrenoids, like chloroplasts are real protoplasmic bodies. It now becomes necessary to state what is meant by Being alive as used above. By that I mean possessing the power to carry on the peculiar synthetic and destructive chemical processes which involve utilization and release of chemical energy. If one includes all the structures that you mention, and adds the cell walls, there is just as much reason for adding crystals of calcium oxalate and other crystalline inclusions, according to which the whole body becomes protoplasm and the term protoplasm absolutely devoid of meaning." This statement was referred to in connection with the statements of Cowles and Chamberlain. In a number of important respects, it differs from all other statements, and, therefore, it is of special interest.

After stating her position, Miss Stokey says "My inclusions and exclusions are based in part on their origin and in part on their persistence or stability. I include in protoplasm the parts of the cell which arise from the division of similar parts and not those which arise *de novo*. I do not include those parts which maintain their identity after the death of the cell or which do not have the unstable structure which is characteristic of cytoplasm and the nucleus,—structures which die." Nothing similar to this very suggestive statement has occurred elsewhere in the correspondence.

Five botanists who replied took a third position which was an outgrowth of the questionnaire, namely that the unit of protoplasm is the protoplast or whole cell content. These botanists are C. E. Allen, C. O. Appleman, T. C. Frye, J. B. Overton, and R. Stratton. Allen says "Both in print and in speech there is a good deal of confusion among biologists as to just what we mean by protoplasm and cytoplasm. We have thrashed over the question here at various times, and have always come back to the definition which—I think, was originally due to Strasburger—namely, that protoplasm includes everything within the wall, and that cytoplasm includes all the protoplasm except the nucleus. In case there is no wall or equivalent non-living structure, the whole substance of the cell is protoplasm. These definitions are at least workable. When we come to the question of just what parts of the protoplasm are living and what are non-living, we find ourselves involved in endless discussion. To attempt, as some

do, to define protoplasm as living matter makes the definition depend in turn upon just what is meant by living matter—a conception which no one at present can define." This strong statement may well stand for the view that the whole protoplast constitutes the protoplasm. In connection with it, the logic of Jeffrey and the statement of Shull about what it means to be alive should be recalled.

Appleman is in general accord with Allen, but the following must be quoted: "The protoplasm of the cell may be conceived to consist of a polyphase colloidal system with systems within systems. The nucleus would be a system within a system. Smaller systems may exist within the nucleus. Each system is bounded by a surface membrane which is an integral part of the system and merely represents local concentration of certain constituents of the system. In the development of this conception I include in protoplasm not only the biocolloids but all other substances dispersed in the dispersion medium, as salts, sugar, starch grains, etc." It seems possible from Appleman's statement regarding membranes that he may regard the cell-wall part of the protoplasm.

C. C. Curtis and R. B. Harvey follow the fourth and last position arising from the questionnaire, namely that the whole cell, including the wall, constitutes the unit of protoplasm. Also it is not certain that Appleman should not be placed here rather than with the last group above. O. F. Curtis was placed in the second group,—but he has strong leanings toward the present one. First he says "Ordinarily I would consider statement number 2, 'Plant protoplasm consists of cytoplasm and nucleus, plus plastids,' perhaps the easiest working basis for distinguishing protoplasm from other portions of the cell." Then, after a statement somewhat similar to Allen's, he says "For this reason I often point out that your statement number 4, 'Plant protoplasm consists of the whole cell content plus the cell wall,' is in many respects less disturbing, for there is then no danger of leaving out any part that may be of great importance in the working of the cell." The two quotations from the writer are from the catagories used at Cincinnati. R. A. Harper referred the writer to his discussion of "The structure of protoplasm" in his presidential address for the Botanical Society of America, at the Pittsburgh meeting, December, 1917. The writer understands that this

paper favors the view of protoplasm considered in this paragraph; but there was no thought of attempting to define protoplasm, except for the purpose of the discussion at hand, and one might well object to being held for a position taken for a special purpose several years ago.

Finally, we have reached a group of botanists whose statements cannot be classified according to the criteria used in this paper. R. C. Benedict, E. A. Bessey, B. M. Duggar, F. E. Lloyd, W. J. V. Osterhout, G. J. Peirce, and H. E. Pulling constitute this group. Benedict regards the idea that cytoplasm and nucleoplasm constitute protoplasm "standard and teachable in elementary courses." He thinks the definition which would add plastids "not worth consideration as distinct from others," presumably because he regards plastids as part of the cytoplasm. He believes also that the protoplast, or this plus the wall, may be regarded the unit of protoplasm. He says these dispositions are "both defensible but require more than elementary courses to develop and be understood." Bessey says in part that he agrees with Sharp on some points and differs on others. Part of his statement runs thus: "I think we must look upon the protoplasm of the cell, not as a series of small biophores floating in a non-living medium, but as a polyphase colloidal system whose chemical and physical inter-actions are the phenomena which we interpret as life phenomena. It would be idle, then, to say that there is only one particular chemical substance in the cell that is alive and that the rest is not alive. It is the sum total of the protein and related substances that contribute to the life phenomena." The statement is no less valuable, simply because the present writer does not attempt to place it in his scheme. Apropos of placing Sharp and not placing Bessey, who agrees in many respects, it may be added that Schaffner, Sharp, Livingston, and one or two others might perhaps have been placed in this galaxy of botanists who seem to be unclassifiable for our present purpose.

Duggar, it appears to the writer, has made a contribution which no doubt reflects the feelings of some others with respect to being classified, and which may be otherwise more valuable because it does not fit into our scheme. He says "It is extremely difficult and often unfortunate, it seems to me, to

commit one's self to categorical distinctions. To the enumeration of four categories designated in your letter the criticism might be made that no one of them would properly define protoplasm at all times; for example, at an early stage of growth a plastid might be protoplasmic, while later, though recognized morphologically as a plastid, it might be made up largely if not entirely of fat or gum-like materials. Since the protoplasm frequently penetrates the cell wall, the cell wall must often be an intimate part of the whole unit, but it may be that this is not always so. In the same way one can work out a close relationship between almost any product of the cell and the cytoplasm or nucleoplasm, and it would be difficult to say just when sufficient 'control' of a product ceased, or when it became less subject to the coordinating properties of the protoplasmic matrix. In short I could not accept any of your four categories in toto."

Peirce replied in terms of colloidal chemistry. His statement was prepared carefully for a forthcoming text book, and we quote it in full as follows: "Protoplasm is a colloidal mass in which water and a great variety of substances exist together, the water being the solvent of many of these substances, the medium of colloidal suspension, and of emulsion of others, thus being external to some and enclosed in others, and therefore held with a firmness which may be overcome only with the application of great force. The various colloidal systems forming the colloidal mass called protoplasm may have only water in common; for they may consist of water and gelatinous substances, sols or gels, the water being external to and suspending the sol or gel and escaping with ease, or enclosed within the gel and removable only on the application of many atmospheres of pressure; of water and proteins, complex compounds with large molecules, which are condensation products of amino-acids, behaving like acids toward strong bases and like bases toward strong acids, since they contain groups, carboxyl and NH^2, giving them these respective properties; and of other substances. Thus we have, in its static condition, an enormously complex mechanical, physical-chemical, physiological system; but since it does not exist in a perfectly static condition, the complexity of the system is added to directly in proportion to the activity of the protoplasm, and

its activity is entirely dependent upon the relative proportions of the actions and reactions of the members, inclusions, and products of its component colloidal systems."

In the letter which accompanied this statement, Peirce says "I am somewhat amused to think of the contrast between this statement and some which you will no doubt receive from others of the sixty to whom you have sent this letter. I can imagine, for example, my colleague, Dr. Campbell, scarcely using a single one of the words which I have employed, and yet his idea of protoplasm is as clear and definite as mine, perhaps more so." The reader is here referred to Campbell's statement in a previous paragraph of this paper. It seems more understandable for the beginner, though no doubt more open to criticism than Peirce's statement.

Pulling has likewise failed to see any great value in the classification used in this paper, but has made a keen analysis of the situation regarding protoplasm. His statement runs thus: "I do not see how it is possible for anyone to subscribe to any of the four statements, because the terms are not the same. Protoplasm is, I think, generally intended as the name of a hypothetical living substance. By hypothetical I merely mean that no substance has ever been indentified as living, but the conviction remains that it will be some day found. When found it must be indentified by how it behaves. Nucleus cytoplasm, plastid, cell wall are all topographical terms. They refer to parts of the cell. How then can one say that protoplasm consists of these things? Of course it may be so. If what the microscopist sees when he looks at cytoplasm is living matter, then it is protoplasm. But all he knows is that what he sees is a portion of the cell in which (by definition of the term protoplasm) there is protoplasm while this portion of the cell exhibits those characteristics that we consider are indicative of life. I do not think this is splitting hairs. A crystal within the cytoplasm is termed an inclusion, so is a fat droplet. What evidence is there that all the visible colloidal material is not also an inclusion? The protoplasm may be an infinitely diluted constituent. I think that we need not hesitate to say that there is protoplasm in nuclei, cytoplasm, plastids, and in all cell walls during part of their life, or existence as part of a living cell, using the term protoplasm as defined above. I don't see that the problem of protoplasm or of life,

or of the functions or characteristics of cell parts is the clearer for such a statement, but I make it in the hope that it will be of service to you." Pulling's statement seems unanswerable, and if, by any chance, protoplasm is the attenuated substance which he thinks it may be, the beliefs of all who have contributed to this paper are wrong. Whatever may be the nature of protoplasm, his arguments, like those of Duggar, strike a hard blow at the method of classification used in this paper and many other current treatments of protoplasm.

Osterhaut states that in a broad sense he used the term protoplasm to include the whole cell content and in a narrower sense he excludes vacuoles, crystals, oil drops, starch grains, etc. He seems at one time to belong to our second group and at another to our third.

Because of lack of space, there are some answers not quoted, which contain matter that would add considerably to the merits of this paper. Among these are the replies of Bartlett, Cook, Harvey, Kern, Lloyd, Pammel, Pool, and Shaffner. Some of the points made in these replies will appear in the summaries below.

Summarizing first on the four points which are the main outgrowth of the questionnaire, seven botanists replied to the effect that protoplasm consists essentially of cytoplasm and nucleoplasm. Thirty-eight would add to this plastids, and some of them one or two other portions of the protoplast. Five would make protoplasm synonymous with the protoplast, or whole cell content. Two or three would add to this the cell wall. Seven sent in replies which cannot be classified according to our scheme. Two heads of departments, J. M. Coulter and E. M. Freeman, referred the writer to other men in their departments.

On other matters partly suggested in the questionnaire, thirteen have referred to protoplasm as the living matter of the cell, and two have stated that it is not possible to distinguish between living and non-living. Ten have emphasized the fact that protoplasm is a polyphase colloidal substance. Six regard vacuoles, with their cell sap, part of the protoplasm, and thirteen have stated that vacuoles should be excluded. Five regard pyrenoids a part of the protoplasm, and three would exclude them. One would make chlorophyll a part of the protoplasm, and five would exclude it. Blepharoplasts,

chondriosomes, and centrosomes are each mentioned by one or two as parts of the protoplasm. Three mention water as a portion of protoplasm, and an equal number would exclude it. Two would include that portion of water which is in intimate relation with other portions of protoplasm and exclude the rest. Protoplasm and cytoplasm are regarded as synonyms by two botanists. These data give some idea of the unsettled condition of opinion on some points not otherwise summarized herein.

An occasional reply mentions food material, mucilage, gums, inulin, sugars, fats, oils, resins, tannin, aleurone, enzymes, acids, crystals, cystoliths, and other miscellaneous substances as not belonging to protoplasm. Of course those who regard the protoplast or the whole cell the protoplasmic unit would include whatever of these might be present in any cell as part of the protoplasm of that cell. However, these substances were uniformly mentioned by persons who take a more limited view of the composition of protoplasm.

Some interesting comments were made respecting the cell wall. Three persons stated that the wall contains protoplasm, and two of these regard the wall partly alive. Martin says "In the more advanced classes, I teach that in some cases at least a portion of the wall is modified cytoplasm still living. There are a number of cases where this is certainly true. I have recently been working on the papillae of the stigma of the Easter Lily, finding that the inner one-half or more of the wall is modified cytoplasm still maintaining life sufficient to modify itself into a mucilaginous substance. Strands of wall and cytoplasm grade into each other."

We have classified as best we could the opinions of a considerable number of botanists regarding the nature of protoplasm, viewed from the standpoint of visible cell contents. We have also considered some replies which do not fit into the system of classification used herein. For the writer, the results would seem worth while, even if he got no more from them than the reactions of many botanists on a problem so important, so difficult, and so little understood. Yet it seems scarcely possible that any botanist, except possibly those who have spent most time on the problem, could study carefully what has been presented without having his ideas regarding

protoplasm modified in some manner and his understanding of this very difficult and unsettled problem enlarged.

While no new idea has been advanced in this paper concerning protoplasm, some very interesting and illuminating statements have been introduced; and we have run the whole possible gamut from the notion that protoplasm is synonymous with cytoplasm to the other extreme which makes the unit of protoplasm coextensive with the cell. Pulling has injected the idea which one finds occasionally that we may not see protoplasm with our microscopes, this substance being perhaps so finely granular as to be ultramicroscopic. In view of diversity of opinion, it is questionable whether we had better say much about protoplasm, and even whether it might not be better to abandon the term and speak and write wholly in terms of cell structure. Chamberlain makes little use of the term, and Harper asked at Cincinnati, perhaps half in jest, whether we might not get on better without using the term protoplasm.

The position that protoplasm consists of cytoplasm and nucleoplasm is very indefinite since cytoplasm may mean anything from the more finely granular material of the cell outside the nucleus to the whole cell content outside the nucleus. It is probable that all those who took this view had the first position in mind; but Miss Stokey has given reasons why the position which would add plastids and some other inclusions which arise from division is much more tenable. This is the position taken by nearly two-thirds of the botanists who replied; and the writer is willing to modify his previous position and accept this tentatively, not because it is the majority opinion, but for reasons stated in this paragraph and elsewhere in this paper. Though accepting it for the time, the writer thinks that a view of protoplasm which stops short of the whole protoplast is so difficult to present to elementary students that he will probably be content to touch protoplasm lightly as the living substance of the cell and give the remainder of the discussions in terms of cell structure. In advanced classes he expects to treat protoplasm as a polyphase colloidal system, and state the various ideas regarding what portions of the cell may or may not be protoplasm. Of course all belongs to the complex colloidal system whether protoplasm or not.

Next in order, we may consider the not commonly accepted position which the writer held when he began this investigation, namely that the unit of protoplasm and the protoplast may be regarded as synonymous. This position rests, at least in part, on the facts that every part of the cell probably contains protoplasm at some time and that we have not yet made a good beginning of ascertaining what is and what is not alive, or what is and what is not protoplasm. The writer very reluctantly recedes from this position which is the most workable one that can be taken and has at the same time strong arguments in its favor. As brought out previously in this paper, the inclusion of the wall in the idea of the unit of protoplasm is the logical position for those who include the whole protoplast.

Two men, Buller and Mottier, were quite insistent at Cincinnati that protoplasm should be passed over as "the physical basis of life" or the living substance of the cell. Biologists can agree on this much. Dismissing the supposition that protoplasm may possibly be an attenuated ultramiscropic substance of which we know nothing, doubtless we will also agree that it is a complex colloidal substance, the chemistry of which is not yet thoroughly known. Beyond this, there is no agreement regarding protoplasm, except that cytoplasm or some portion of cytoplasm is protoplasm. A careful analysis of opinions of botanists quoted in this paper brings one to this peculiar conclusion. Consequently, no more inclusive position with respect to the physical content of protoplasm can be assumed certainly; and we should take our positions with great respect for the opinions of others.

How long we may have to continue in this uncertain state of mind respecting protoplasm is wholly problematical. Though most of our ideas of protoplasm are at present quite hypothetical, biologists and chemists have accomplished a good deal in this very difficult field of research. With further study, it seems probable that the fragments of knowledge may be rewoven from time to time until the woof of knowledge regarding protoplasm may be fairly complete.

In the present state of knowledge of protoplasm, the matter of prime importance is that we should not feel so certain of any position taken as to attempt to treat as established that which is quite uncertain. The few well established facts may

be stated with confidence. But the many points that are purely hypothetical should be handled in research and in teaching without attempting to hold any one position to the exclusion of the possibility of another one being preferable. If, then, rather than attempting the impossible in way of reaching some positive position, this paper may contribute to open mindedness respecting protoplasm, and suggest treatment commensurate with present knowledge, it will have accomplished the best possible service.

Finally, the writer wishes to express his very great appreciation of the aid given so generously by busy workers.